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Agreee: To ligate the purified vector + insect and transfect with appropriate controls.

Infection Rx:

Vector ~ 100ng/ μ l	=	1 μ l	1 : 1 "	1 Vector alone
Insect ~ 25ng/ μ l	=	4 μ l	"	2 " + real cyto
5x buffer	=	4		3 " any purified
Ligase T4		1		3 - Tag 1.5
H ₂ O		10		4 " "
		20 μ l	at 25°	5 " + DV 1.5
			3 hrs.	6 " "

transformation using DH5α Max eff. cells.

that produced used 2.5 μ g lig alone Mix / transformation
final volume after adding SOC - 500 ml.

Plated 25, 50 & 100 μ l of each,

Control diluted to 1:10 and plated 25, 50 & 100 μ l.
Normal transformation efficiency
Vector only - few blues because of the contamination $\sim 1.5 \times 10^9$
no whites.

Vector + rest - ligated w/o any purification, lots + lots of colonies
transformation quite efficient.

Tag + DV } 2 mM - didn't work

Tag alone Mg

Tag " } 1.5 mM

Tag + DV Mg

- all purified gave low. efficiency of transformation compared to
un. " - Vector + rest

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Signed & Understood by m ,

Date

Invented by

Date

14/12/94

Recorded by

12/13/94

R. Subramanian